

We claim:

1. A method for estimating the effects of a plurality of factors and at least one of a plurality of interactions between the factors in a gene expression microarray experiment generating a microarray dataset wherein the factors include a gene factor and at least one non-gene factor and the interactions include at least one gene interaction, the gene factor being orthogonal to the other factors, the method comprising the steps of:
 - (a) estimating the factor effects based on a plurality of averages of the microarray dataset; and
 - (b) estimating the at least one gene interaction effects based on a plurality of averages of the microarray dataset and on the estimated factor effects from step (a).
2. The method of claim 1 wherein each non-gene factor is characterized by a number of levels, the step of estimating the main effects includes inverting a square matrix of size p wherein p is equal to the sum of the number of levels for each non-gene factor minus the number of non-gene factors.
3. The method of claim 1 wherein each factor is characterized by a level, the step of estimating the interaction effects includes inverting a square matrix of size p' wherein p' is equal to the sum of the number of levels for each non-gene factor minus the number of non-gene factors minus one.
4. The method of claim 3 wherein the non-gene factor includes a variety factor and the step of estimating the interaction effects further includes estimating the variety-gene interaction effects for each gene based on the inverted square matrix of size p' .
5. The method of claim 1 wherein the non-gene factors includes a variety factor.
6. The method of claim 1 wherein the non-gene factors includes an array factor.
7. The method of claim 1 wherein the non-gene factors includes a dye factor.

8. The method of claim 7 wherein the dye factor has two levels.
9. The method of claim 6 wherein the array factor has two levels.
10. The method of claim 8 wherein the array factor is balanced with respect to the dye factor.

5 11. A method for estimating at least one gene-variety interaction in a gene expression microarray experiment having an experimental design characterized by a number of degrees of freedom, q , and defined by a gene factor, a plurality of non-gene factors, a plurality of two-factor interactions wherein a full replication of genes is present for every combination of the plurality of non-gene factors, the method comprising the steps of:

- (a) inverting a first square matrix characterized by a size, p , wherein $p < q$;
- (b) estimating at least one of a plurality of non-gene factor effect from the first square matrix inverse;
- (c) constructing a second square matrix based in part on the estimated non-gene factor, the second square matrix characterized by size, p' , wherein $p' < q$;
- (d) inverting a second square matrix; and
- (e) estimating at least one gene-variety interaction from the inverted second square matrix.

20 12. A method for estimating at least one gene-variety interaction in a gene expression microarray experiment generating a dataset and having a design characterized by a arrays, v varieties, n genes, and d dyes wherein a full replication of genes is present for every combination of arrays, varieties and dyes, the method comprising the steps of:

- 25 (a) constructing a global data vector, \mathbf{d} , based on a plurality of averages of the dataset;
- (b) constructing a square matrix, \mathbf{T} , characterized by a size, p , wherein $p = a + v + d - 3$;
- (c) inverting the square matrix, \mathbf{T} ;
- 30 (d) estimating the global effects, $\boldsymbol{\tau}$, wherein $\boldsymbol{\tau} = \mathbf{T} \mathbf{d}$;

- (e) constructing a square matrix, T_g , characterized by a size, p' , wherein $p' = p - 1$;
- (f) constructing a gene-specific data vector, d_g , based on a plurality of averages of the dataset;
- 5 (g) inverting the square matrix, T_g ; and
- (g) estimating the gene-variety interaction, τ_g , wherein $\tau_g = T_g d_g$.

13. A system for estimating the effects of a plurality of factors and at least one of a plurality of interactions between the factors in a gene expression microarray experiment generating a microarray dataset wherein the factors include a gene factor and a variety factor and the interactions include a variety-gene interaction, the gene factor being orthogonal to the other factors, the system comprising:

- 10 (a) a processor;
- (b) a memory in signal communication with the processor;
- 15 (c) a program stored in the memory, the program capable of being executed by the processor, the program including the steps of:
- (i) estimating the main effects based on a plurality of averages of the microarray dataset; and
- (ii) estimating the interaction effects based on a plurality of averages of the microarray dataset and on the estimated factor effects from step (i).

20 14. The system of claim 13 wherein each factor is characterized by a level, the step of estimating the main effects includes inverting a square matrix of size p wherein p is equal to the sum of the levels for each non-gene factor minus the number of non-gene factors.

25 15. The system of claim 13 wherein each factor is characterized by a level, the step of estimating the interaction effects includes inverting a square matrix of size p' wherein p' is equal to the sum of the levels for each non-gene factor minus the number of non-gene factors minus one.

30 16. The system of claim 15 wherein the step of estimating the interaction effects further includes estimating the variety-gene interaction effects for each gene based on the inverted square matrix of size p' .

17. The system of claim 13 wherein the non-gene factors includes a variety factor.

18. The system of claim 13 wherein the non-gene factors includes an array factor.

19. The system of claim 13 wherein the non-gene factors includes a dye factor.

20. The system of claim 19 wherein the dye factor has two levels.

5 21. The system of claim 18 wherein the array factor has two levels.

22. The system of claim 20 wherein the array factor is balanced with respect to the dye factor.

23. The method of claim 1 wherein the interactions further include an array-dye interaction.

10 24. The system of claim 13 wherein the interactions further include an array-dye interaction.

15 25. A method for estimating the effects of a plurality of factors and at least one of a plurality of interactions between the factors in a gene expression microarray experiment generating a microarray dataset wherein the factors include a gene factor and a plurality of non-gene factors and the interactions include at least one of a gene-non-gene interaction, the gene factor being orthogonal to the non-gene factors, the method comprising the steps of:

(a) constructing a first data model including only non-gene factors and non-gene interactions;

20 (b) estimating the effects of the non-gene factors and non-gene interactions based on the first data model and on a plurality of averages of the microarray dataset;

(b) creating a transformed dataset from the microarray dataset and the factor and interaction effects estimated in step (a);

25 (c) constructing a second data model including the gene factors and the gene interactions; and

(d) estimating the gene-non-gene interaction effects based on the second data model and a plurality of averages of the transformed dataset.

26. The method of claim 25 wherein the non-gene factors includes a variety factor.

27. The method of claim 26 wherein the non-gene factors includes an array factor.

5 28. The method of claim 27 wherein the non-gene factors includes a dye factor.

29. The method of claim 28 wherein the dye factor has two levels.

30. The method of claim 27 wherein the array factor has two levels.

31. The method of claim 30 wherein the array factor is balanced with respect to the dye factor.

32. The method of claim 28 wherein a non-gene interaction includes an array-dye interaction.

33. The method of claim 32 wherein the transformed dataset is created according to the equation:

$$x_{ijkgs} = y_{ijkgs} - \hat{\mu} - \hat{A}_i - \hat{D}_j - (AD)_{ij}$$

where

x_{ijkgs} is the transformed measurement of y_{ijkgs} measurement,

y_{ijkgs} is the $ijkgs^{\text{th}}$ measurement;

$\hat{\mu}$ is the estimated mean of all measurements;

\hat{A}_i is the estimated array effect for the i^{th} array;

20 \hat{D}_j is the estimated dye effect for the j^{th} dye;

$(AD)_{ij}$ is the estimated array-dye interaction effect of the i^{th} array and the j^{th} dye.

34. A computer readable media comprising instructions encoded therein for a computer to perform the method of claim 1.